

acres were made from smaller subplots selected at random, or were made by collecting all *Lychnis* flowers encountered along lines walked edge-to-edge or corner-to-corner of a field.

The number of styles was counted for every flower collected. Each collection was tabulated, and cumulative records were kept, separately, for each plot from which repeated collections were taken. All ovaries with an atypical number of styles were preserved in formol-acetic-acid-alcohol as evidence of collection and for histological study.

Ovaries of most (66.9 percent) of the 21,669 flowers examined bore the typical five styles; style numbers for the remainder (33.1 percent) ranged as follows: 0, 1, 2, 3, 4, 6, 7, 8, 9, and 10 styles per ovary (Table 1). The number of carpels, placentae, and placental vascular strands of a given ovary, determined from paraffin sections, varied directly with the number of styles it bore, except for styleless ovaries and those with certain other irregularities.

Of 18 styleless ovaries sectioned, carpels ranging in number from three to seven, inclusive, were found. Similarly, the number of styles of a given ovary did not correspond always to the actual number of carpels. For example, ovaries with one to four styles, inclusive, could be expected to have, correspondingly, one to four carpels. However, any one of these might actually have five to eight (possibly more) carpels; this suggested that one or more styles of a particular ovary had failed to develop.

The typical, young, five-styled ovary of *Lychnis alba* has axile placentation with five carpellary septa connecting the ovary wall to the columnar placental region. Subsequently these septa break, and the whole central region (actually containing five placentae) thus detached

from the ovary wall becomes the free-central placenta.

Except for stated irregularities, one-styled ovaries have one carpel, one locule, and one marginal placenta. Two-styled ovaries have two carpels and two locules, the placenta becoming free-central when the two oppositely placed carpellary septa break. Ovaries with three to ten styles, inclusive, develop typically.

The placental column of a typical five-styled, five-carpellate ovary has five prominent, equally spaced, radially prolonged vascular strands (appearing starlike in placenta cross sections) extending platelike throughout its length. These vascular strands vary directly with the number of carpels and placentae but not always with the number of styles of a given ovary.

Ovaries of *Lychnis alba* may have more carpels, placentae, and vascular strands than styles, but the reverse situation was not found.

Significant variations in style number appeared in every larger collection of *Lychnis alba* flowers examined during 1958; smaller collections sometimes produced fewer variations, most (and occasionally all) ovaries bearing five styles. Twenty-nine individual plants observed daily in a greenhouse throughout their entire flowering period from October 1958 through January 1959 exhibited a similar variance (8). Twenty-five of these produced ovaries bearing significant variations in style number (four to nine, inclusive); one was an androhermaphrodite; in one most ovaries bore five styles; and in two all ovaries bore five styles, strictly.

In this respect, one plot of *Lychnis alba* (West Liberty plot No. 5) proved especially interesting because the most complete range of variation in style

numbers found in any one plot studied during 1958 (zero to ten, inclusive) occurred in the 4142 flowers collected from it. Two hundred and forty-one gynohermaphrodite flowers with styles ranging in number from three to eight, inclusive, were likewise found in this plot.

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9 February 1959

Incidence of Sex Chromatin in *Gallus domesticus*

Abstract. The presence of so-called sex chromatin has been demonstrated in the interphase nuclei of the cells of the domestic chicken (*Gallus domesticus*). Definite sex dimorphism was observed for the incidence of this nuclear component; the frequency of its occurrence in females was at least ten times that of its occurrence in the males, ranging from 22 percent in the duodenal muscle cells to 52 percent in the epidermal cells of a growing feather.

Considerable corroborative evidence for the existence of sex chromatin in mammals has been accumulating since Barr and Bertram (1) first drew attention to the "nuclear satellite" and its possible usefulness in establishing the genetic sex of an individual. The term *nuclear satellite* was subsequently (2) changed to *sex chromatin*. All the reports to date, of which those of Cantwell *et al.* (3) and of Osuchowska and Suminski (4) are among the latest, clearly indicate the sex-dimorphic nature of the distribution of sex chromatin. In mammals, it is the cells of genetic females that are richly provided with this strongly basophilic, Feulgen-positive material. In males, sex chromatin is either nonexistent or occurs only infrequently. One is not surprised, therefore, to find a generalization, suggested by Prince *et al.* (5), that sex chromatin is the heterochromatic portion of the 2 X chromosomes. More recently Klinger (6) came to the same conclusion. It should be recalled in this connection

Table 1. Data indicating the total number of *Lychnis alba* flowers collected from each general area, the spread of variations in style number, the number of examples and totals for each category of variation, and the grand total of all flowers examined during 1958.

No. of styles	No. of flowers			Total No. of flowers of given style No. from the three areas
	Iowa greenhouse, 19 May–23 June (7)	Iowa outdoors, 20 June–23 July	Minnesota outdoors, 1–27 Aug.	
0	0	8	24	32
1	0	13	14	27
2	0	13	31	44
3	4	46	88	138
4	149	218	391	758
5	2,607	4,967	6,933	14,507
6	317	1,766	2,454	4,537
7	64	661	712	1,437
8	5	90	68	163
9	1	13	10	24
10	0	2	0	2
Totals	3,147	7,797	10,725	21,669

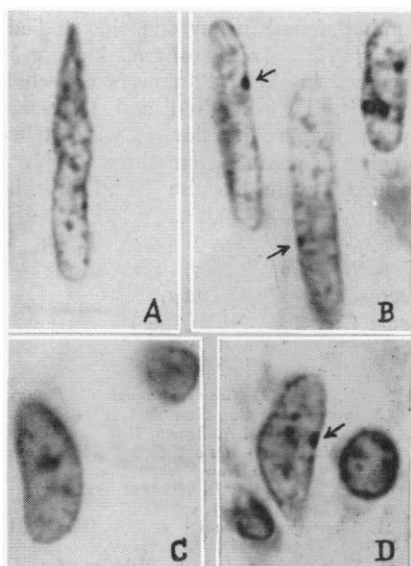


Fig. 1. Representative nuclei from the epidermis of a growing feather and muscle layer of the duodenum. A and C, from males; B and D, from females. Arrows point to sex chromatin.

that Smith, as early as 1945, presented evidence (7) showing that female sex in *Archips fumiferana*, a lepidopteran, could be identified on the basis of heteropycnosis in the intestine. He concluded that these heteropycnotic bodies are sex chromosomes—a somewhat puzzling deduction in view of the fact that in lepidoptera females are heterogametic. Thus, one would expect a still greater degree of heteropycnosis in the lepidopteran males (that is, if his postulate was correct), a situation contrary to the one observed by Smith.

The presently reported study (8) was undertaken to (i) examine the phenomenon of sex chromatin in relation to class Aves, in which the female sex is heterogametic (a situation just the reverse of that in mammals), and (ii), in case the results were positive, to use sex chromatin as a tool in studying the genetic sex of rather frequently occurring sex intergrades in the domestic poultry. On the basis of the interpretation of the published mammalian data, the expectation was that, if sex chromatin occurs in birds, it would be found to be characteristic, in the main, of quiescent nuclei in the male sex.

The study involved 3-week-old New Hampshire chickens. Specimens for histologic examination were obtained from the following organs and tissues: brain, spinal cord, liver, intestine, pancreas, gonads, skin, and feathers. The specimens were fixed in a mixture of formal-acetic-acid-alcohol (40 percent formaldehyde, glacial acetic acid, 95-percent EtOH, and distilled water in the ratio of 2:1:3:3, respectively), and stained with Harris' hematoxylin.

Of the tissues studied, smooth muscles in the duodenum and dermal and epidermal components at the base of a growing feather were found to be best for demonstrating the presence of sex chromatin in the chicken (Fig. 1). While there were individual differences among the birds in the incidence of sex chromatin, clear-cut dimorphism was unmistakable: in females, the sex chromatin was much more prevalent than in males (Table 1).

The duodenal and feather preparations also were subjected to Feulgen reagent as well as to ribonuclease digestion, followed by the Unna pyronin-methyl-green staining procedure. Both of these histochemical tests indicated the presence of deoxyribonucleic acid in the sex chromatin, a point which had been demonstrated earlier for the mammalian material (9). This fact, together with the general morphology of sex chromatin in the chicken (its extreme basophilia and its occurrence only in the interphase nucleus where it is closely apposed to the nuclear membrane) led us to conclude that it is analogous to the sex chromatin of mammals.

If this point of view is accepted, then it makes untenable the suggestion (1, 5, 10) that sex chromatin is the heterochromatin of the two sex chromosomes; the somatic nuclei of the female chicken carry only a single sex chromosome.

On the basis of the observations reported above, we agree with the view of Witschi (11) that sex chromatin is not necessarily related to X chromosomes, although it is, clearly, an incompletely-sex-limited trait characterizing the female sex. Obviously, this generalization at present can only be applied to the mammalian and avian species thus far studied. The elaboration and possible modification of the concept, together

Table 1. Frequencies (percentages) of occurrence of cells with identifiable sex chromatin.

Bird speci- men No.	Feathers		Duodenal muscles
	Dermal	Epi- dermal	
<i>Female</i>			
1	44	51	28
2	40	46	21
3	39	55	15
4	31	52	22
5	36	56	22
Av.	35	52	22
Total No. of cells	1100	931	732
<i>Male</i>			
1	1.6	3.5	4.7
2	4.8	5.4	3.8
3	1.1	2.1	2.9
4	0.4	5.2	1.2
5	1.2	1.0	0.6
Av.	1.9	3.4	2.6
Total No. of cells	938	946	782

with elucidation of the basic nature of sex chromatin, will have to await further studies.

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16 February 1959

